than targeting either alone. Erythropoietin-producing hepatocellular carcinoma-A2 (EphA2)-specific CAR T cells were used to target the A549 tumor cells. EphA2-specific T cells when administered together with FAP-specific T cells, resulted in a significant decrease in tumor growth and increased survival compared to mice that received either EphA2- or FAP-specific T cells alone. Our study underscores the value of co-targeting both CAFs and cancer cells to increase the benefits of T-cell immunotherapy for solid tumors.

POSTER SESSION 2: STEM CELL BIOLOGY

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Fully Automated Clinical-Scale Separation of CD133$^+$ Cells From Bone Marrow Aspirate

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There is a growing interest in CD133 antigen expressing stem cell in the field of regenerative medicine such as cardiovascular, peripheral artery, and liver disease. Investigators in particular concentrate on bone marrow-derived stem cells for these applications. Today the clinical scale enrichment of CD133$^+$ cells has to be performed as a complex procedure involving numerous manual handling steps.

We have developed a fully automated clinical scale process within a closed sterile system to purify CD133$^+$ cells from human bone marrow aspirates. In this context, erythrocyte reduction, generation of autologous plasma, labeling time and the conditions for immunomagnetic separation were optimized.

To determine the process performance, CD133$^+$ cells were separated from human bone marrow aspirates with an initial volume of about 60 mL (n=10). We performed colony-forming unit (CFU) assays, which allowed us to evaluate the differentiation potential of the enriched cells.

The total processing time was reduced from about 4.5 h (previous manual process) to 2.5 h. The number of enriched CD133$^+$ cells was 7.9x10^5 (range: 3.7x10^5 to 1.9x10^6). The average yield was 47% and the average viability of the separated CD133$^+$ cells achieved 90% (range: 69.9% to 96.9%). The depletion of CD133 negative cells was >99.9%. CFU assays perfomed after the fully automated enrichment process showed that the CD133$^+$ cell fraction contained primitive and multipotent progenitor cells, such as CFU-GEMM and CFU-GM. The cell separation system described provides a safe and easy way to purify CD133$^+$ cells from bone marrow aspirates within 2.5 h without any intermediate manual steps. The cell preparation in a closed sterile system facilitates a fast and robust enrichment of CD133$^+$ cells. The cells are eluted in a small volume (6 mL) and can be used directly for further applications according to requirements e.g. for use in regenerative medicine.

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Promotion of Wound Healing by Cord Blood Derived Unrestricted Somatic Stem Cells (USSCs) in a Murine Wound Healing Model and Analysis on Their Bio-Distribution by In Vivo Bioluminescent Imaging (BLI)

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Background: Delayed healing of skin wounds is a major morbidity. Repeated wounding is characteristic in patients with recessive dystrophic epidermolysis bullosa (RDEB), caused by mutations in *COL7A1* gene. Stem cell therapy offers an option in treating this disease (Kiru/Cairo et al. PNAS, 2011). Recently, cord blood (CB) derived pluripotent stem cells, USSCs, have been applied in several animal models of degenerative diseases with beneficial outcomes.

Goal: To determine the potential of USSCs in the treatment of RDEB and its associated wounding phenotype.

Method: CB-USSCs were characterized for genetic and functional properties. Their in vivo functions were evaluated in a murine full-thickness excisional wound healing model and by bioluminescent imaging (BLI), using USSCs modified with a luciferase reporter gene.

Results: CB-USSCs share several embryonic stem cell properties and could be induced to express hallmark genes of keratinocyte differentiation. USSCs constitutively express Col7A1, supporting their therapeutic potential in the treatment of patients with RDEB. In the wounding model, a single USSC intradermal injection promoted epithelialization and facilitated formation and remodeling of epidermis, accompanied by a significantly accelerated rate of wound healing on days 6-10 post wounding ($F_{(1,168)}$=50.8 P < .01). In vivo BLI revealed specific migration of USSCs from a distant intradermal injection site toward the wound, as well as following systemic injection. Temporal quantification on the total bioluminescence indicated an overall 59.9% signal loss over 3 days followed by a 95.06% loss at 1 week. The bioluminescence in the area of wound was then maintained at ~0.5-1% level till the end of the experiment (3 month). USSCs express several chemokine receptors that may mediate their migration to the wound, including CXCR4 (for SDF1), CCR7 (for CCL21) and PDGFRα (for HMGB1). In vitro chemotaxis assays indicated that SDF-1 significantly enhanced USSC migration at a concentration of 100ng/ml, while neither CCL21 nor HMGB1 showed significance even at a concentration of 10μg/ml. The effects of such chemokine/receptor interactions on USSC recruitment in vivo are now being investigated.

Conclusion: These results suggest significant beneficial effects of CB-USSCs on wound healing and raised the possibility of USSC's therapeutic beneifit in the treatment of patients with RDEB.

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Another Method for Thawing Hematopoietic Stem Cells and its Impact in the Recovery of the Transplanted Hematological Patient

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Introduction: There are several hematopoietic-stem cells (HSCs) thawing methods for bone marrow reconstitution. They intend to avoid cell death and patient's side effects due to the dimethyl sulfoxide (DMSO). We propose another thawing method that diminishes cell death and therefore a more rapid hematological recovery.

Material and Methods: The standard thawing-removing DMSO method for cord blood units was described by